

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 2

Amendments to the Claims

This listing of claims will replace all prior versions and listings of the claims in the application.

Claims 1-122 (Cancelled).

--123. (Currently amended) A composition which comprises:

- a) a conjugate ~~comprising of~~ (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) Keyhole Limpet Hemocyanin, an immunogenic protein-based carrier;
- b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and
- c) a pharmaceutically acceptable carrier;

wherein the amount of the conjugated ganglioside derivative is an amount between about 1 µg and about 200 µg, the amount of the saponin is an amount between about 10 µg and about 200 µg, and when the ganglioside is GM2, the GM2:Keyhole Limpet Hemocyanin molar ratio is from 200:1 to 1400:1, the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an antibody to whichever ganglioside is present as a derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GM2, GD2, GD3 lactone, O-acetyl GD3 and GT3; and

~~wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and~~

wherein in the conjugate the ganglioside derivative is covalently bound

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 3

to the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin. --

--124. (Previously presented) The composition of claim 123, wherein the saponin is QS-21. --

Claims 125-129 (Cancelled).

--130. (Currently amended) The composition of claim ~~[[129]]~~ 123, wherein the amount of the saponin is about 100 μ g. --

--131. (Currently amended) The composition of claim ~~[[129]]~~ 123 wherein the amount of the saponin is about 200 μ g. --

--132. (Currently amended) The ~~[[A]]~~ composition of claim 123 which comprises:

- a) a conjugate ~~comprising of~~ (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) Keyhole Limpet Hemocyanin, an immunogenic protein-based carrier;
- b) QS-21, a saponin derivable from the bark of a Quillaja saponaria Molina tree, ~~wherein the saponin is QS-21~~; and
- c) a pharmaceutically acceptable carrier;

wherein the conjugated ganglioside derivative is present in an amount of between about ~~10 μ g and about 50 μ g~~ 1 μ g and about 200 μ g, and the amount of the saponin is about 100 μ g, and when the ganglioside is GM2, the GM2:Keyhole Limpet Hemocyanin molar ratio is from 200:1 to 1400:1, and wherein the relative amounts of such conjugate and such saponin is effective to stimulate or enhance production in a subject of an antibody to GM2, GD2, GD3 and GT3, whichever ganglioside is present as a

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 4

derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GM2, GD2, GD3 lactone, O-acetyl GD3 and GT3; and

~~wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin, and~~

wherein in the conjugate the ganglioside derivative is covalently bound to the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin. --

--133. (Currently amended) A method of treating a subject afflicted with melanoma which comprises administering to said subject an amount of a composition of claim 132 effective to stimulate or enhance production of an antibody to at least one ganglioside selected from the group consisting of GM2, GD2, GD3 lactone, O-acetyl GD3 and GT3 and to thereby treat said melanoma in said subject. --

--134. (Currently amended) A method of stimulating or enhancing production of an antibody to GM2, GD2, GD3 and GT3 in a subject which comprises administering to the subject an effective amount of a composition which comprises:

- a) a conjugate ~~comprising of~~ comprising of (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) Keyhole Limpet Hemocyanin, an immunogenic protein-based carrier;

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 5

b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and

c) a pharmaceutically acceptable carrier;

wherein the amount of the conjugated ganglioside derivative is an amount between about 1 µg and about 200 µg, the amount of the saponin is an amount between about 10 µg and about 200 µg, and when the ganglioside is GM2, the GM2:Keyhole Limpet Hemocyanin molar ratio is from 200:1 to 1400:1, the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an antibody to GM2, GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GM2, GD2, GD3 lactone, O-acetyl GD3 and GT3; and

~~wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and~~

wherein in the conjugate the ganglioside derivative is covalently bound to the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ε-aminolysyl group of the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin so as to thereby stimulate or enhance production of the antibody to GM2, GD2, GD3 and GT3 in the subject, whichever ganglioside is present as a derivative in the conjugate. --

--135. (Currently amended) A method of treating a human subject having cancer ~~cancer in a subject~~ which comprises administering to the subject an effective ~~cancer-treating~~ amount of a composition which comprises:

a) a conjugate ~~comprising~~ of (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 6

- ceramide portion comprising an altered sphingosine base and (ii) Keyhole Limpet Hemocyanin, an immunogenic protein-based carrier;
- b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and
- c) a pharmaceutically acceptable carrier;

wherein the amount of the conjugated ganglioside derivative is an amount between about 1 µg and about 200 µg, the amount of the saponin is an amount of between about 10 µg and about 200 µg, and when the ganglioside is GM2, the GM2:Keyhole Limpet Hemocyanin molar ratio is from 200:1 to 1400:1, the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an antibody to GM2, GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate;

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GM2, GD2, GD3 lactone, O-acetyl GD3 and GT3; and

~~wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and~~

wherein in the conjugate the ganglioside derivative is covalently bound to the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ε-aminolysyl group of the ~~immunogenic protein-based carrier~~ Keyhole Limpet Hemocyanin, so as to thereby stimulate or enhance production of the antibody to GM2, GD2, GD3 and GT3 in the subject, whichever ganglioside is present as a derivative in the conjugate. --

--136. (Previously presented) The method of claim 135, wherein the cancer

Applicants : Philip Livingston and Friedhelm Helling
U.S. Serial No. : 08/477,147
Filed : June 7, 1995
Page 7

is of epithelial origin. --

--137. (Previously presented) The method of claim 135, wherein the cancer is of neuroectodermal origin. --

--138. (Previously presented) The method of claim 137, wherein the cancer of neuroectodermal origin is a melanoma. --

--139. (Previously presented) The method of claim 134 or 135, wherein the administering is effected at two or more sites. --

--140. (Previously presented) The method of claim 139, wherein the administering is effected at three sites. --

--141. (Previously presented) The method of claim 134 or 135, wherein the composition is administered subcutaneously to said subject. --

--142. (Previously presented) The method of claim 141, wherein the composition is administered to said subject at two-week intervals. --

--143. (Previously presented) The method of claim 141, wherein the composition is administered to said subject at weekly intervals. --

--144. (Previously presented) The method of claim 134 or 135, wherein the composition to be administered is prepared prior to administration to the subject by mixing the conjugate and the saponin. --

--145. (Previously presented) The method of claim 144, wherein the conjugate and the saponin are mixed on the day of administration to the subject. -

Claim 146 (Cancelled).